

URINARY TESTS

IN PAPER AND IN TABLET FORM.

Convenient, Cleanly and Competent Methods for Bedside Urinalysis.

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Urinary Test Papers.

A happy suggestion was made some months ago by Dr. G. Oliver, of Harrogate, Eng., that slips of paper might be impregnated with the various reagents employed by the physician in examining the urine, and that he might thus compress into the compass of a vest-pocket case all the reagents likely to be of service in such clinical examinations. Such a pocket case is here offered, and we are confident that it will meet with a hearty welcome from the multitude of those who are willing to derive all possible assistance from simple chemical tests, but who have not the time to fit up for themselves a complete chemical laboratory. This little case contains, first, the indispensable litmus paper—a neutral shade, serving as a test either for acidity or alkalinity of the secretion.

TESTS FOR SUGAR.

The series of test papers includes in the second place these reagents for glucose.

1. Picric acid, with sodium carbonate. This test, introduced by Dr. Geo. Johnson, serves to detect very minute quantities of sugar. In applying the test it is necessary to use a larger quantity of the sodium carbonate than is contained in a single soda paper. Several of the papers may be used, but it is better to carry in the medicine case a supply of the alkali, sodium or potassium carbonate. To make the test, put into the test tube one of the picric acid papers, with 10 minims of the urine, add 20 minims of water and about three grains of sodium carbonate and boil 60 seconds if necessary. If sugar is present the urine assumes a dark red or even a brown color. Even normal urine shows a darkening of color, however, and experiments should be made with normal specimens to determine the amount of change to be expected. If there is more than a trace of sugar in the urine, a smaller quantity will show the reaction, and the experiment may be repeated with 5, 3, and 1 drop successively, and if the reaction is still obtained the urine may be diluted, two, four and eight times with water, and one drop of the diluted fluid tested. In this way a closely approximate estimation may be made of the amount of sugar present, based on the observation that one drop of a fluid containing one grain of glucose to the fluid ounce will show a distinct reaction.

2. Indigo carmine. This is the most sensitive, and by far the best of the tests for sugar.

Place in a test tube 30 minims of water with indigo and a soda paper. Heat the contents of the tube gently until the indigo is dissolved, Then add (best from a pipette) one drop of the urine to be tested, and keep the fluid at the boiling point, without allowing it, however to boil, for sixty seconds. If no effect is produced, add a second drop of the urine and heat once more, and so continue until five drops of the urine have been added. Use no more of the indigo paper than is required to produce a pale blue solution. A portion only of one of the test papers will suffice.

If any notable amount of sugar is present, one or at least two drops will suffice to bring about the reaction. The fluid will change from pure blue to amethyst, then to purple and red, and then will fade to a pale yellow. If the quantity of sugar is very small the color will change only to a purple or red, and in nearly every case five drops of a normal urine will produce this change. If one drop of the urine produce a strong reaction, dilute the urine to $\frac{1}{2}$, $\frac{1}{4}$, $\frac{1}{8}$, $\frac{1}{16}$, etc., in succession, until a single drop ceases to produce a visible change, and estimate roughly in this manner the quantity of sugar present.

In regard to the comparative value of tests for sugar, it may be said that the copper test is the least trustworthy. Among the normal constituents of the urine, uric acid is capable of reducing copper compounds, and numerous substances which may be accidentally present have a similar action. Kreatine and many other organic substances prevent or retard the precipitation of small quantities of cuprous oxide, so that urine containing less than 1 grain of sugar to the fluid ounce, often fails to respond to the copper test, or gives an indication only after half an hour, or even a longer time, and the test is of no value for quantities of sugar less than this. Picric acid forms a valuable indication of the presence of traces of sugar, but a larger amount of alkali must be employed in making the test than can be conveniently put in the form of a test paper, so that a supplementary supply of the alkali must be carried if the test is to be used at the bedside. The reagent is not affected by uric acid, nor by most of the substances constantly or occasionally present in the urine, which reduce copper. It does react, however, with kreatinine, one of the constituents of normal urine, and also with ferrous salts, tannin and inosite, all of which are sometimes found in that secretion. Normal urine, in fact, produces about the same amount of change in color as does a solution of pure glucose containing one-half grain to the ounce. Indigo is capable of detecting a smaller quantity of sugar in the urine than either of the other reagents. One drop of a solution of glucose containing $\frac{1}{2}$ grain to the fluidounce shows a distinct reaction. Of the normal constituents of the urine none of those present in considerable quantity affect indigo. Inosite, which is perhaps always present in minute quantity, reacts like glucose, being distinguished from it by its behavior with the copper test. Of the possible accidental constituents of the urine, only ferrous salts and tannic acid affect indigo, so that we may regard this test as not only the most sensitive by far of any yet proposed, but as practically free from fallacies. Finally, unlike the copper test, this reagent can be kept unchanged for years, especially in the convenient form of these test papers.

ALBUMEN TESTS.

As tests for albumen we furnish in this series the following.

1. Potassio mercuric iodide.
2. Sodium tungstate.
3. Potassium ferrocyanide.
4. Picric acid.

These reagents are all used in connection with citric acid. The test is best made by dropping into the tube containing the urine (30 minims) first a citric acid paper, and allowing a few moments for the acid to become dissolved. If a cloudiness is produced by the acid it is due to the presence either of uric acid or of mucin, or rarely of oleo resins, as in cases where balsam copaiba has been taken medicinally. The urates disappear on warming the urine. Mucin remains, however, and is distinguished from any other constituent of the urine by this behavior. The oleoresinous precipitate is cleared up by boiling, but quickly returns while the urine is still warm.

After observing the effect of the acid alone, add the albumen precipitant, one of the four that have been named. As the reagent dissolves, albumen, if present, is precipitated in the form of a distinct cloud, which is rather increased than diminished on the application of heat. In regard to the individual reagents it may be said that the most sensitive of all is probably the mercuric test paper, which, according to Dr. Oliver, will detect one part of albumen in 20,000 of fluid. Scarcely inferior, however, are sodium tungstate and picric acid, while potassium ferrocyanide, although decidedly less sensitive, is capable of indicating one part of albumen in 10,000 to 12,000. It is therefore not superior to nitric acid, except in convenience — an important, advantage, however.

Of these reagents, potassio-mercuric iodide and picric acid precipitate quinine and other alkaloids, which may be present in considerable quantity in the urine. The precipitates are readily distinguished from albumen, however, by the application of heat, which dissipates them. Alcohol also dissolves them.

All these reagents except ferrocyanide of potassium precipitate peptones, which may be present in the urine. A moderate heat clears up the solution, but on cooling it becomes cloudy again.

Of all the tests ferrocyanide of potassium may be selected for the close resemblance in its indications to those of nitric acid, while for detecting mere traces of albumen, the mercuric salt or sodium tungstate must have the preference, and the results

are conclusive if the precaution be taken to supplement the test in every case by the application of heat.

It may happen that when a very large quantity of albumen is present the paper may become coated with the precipitate which will form instantaneously when it is introduced into the fluid. Although such a contingency must be of very rare occurrence, it is well to remember the possibility; the subsequent application of heat can hardly fail to reveal the presence of the albumen in such a case.

To form an idea of the quantity of albumen present, one has only to observe the density of the precipitate formed when the test is applied in the ordinary way and then to dilute the urine until the reaction becomes just perceptible when the mercuric papers are used. Suppose it be necessary to dilute the urine to 1 200 before this point is reached, we reason that since the reagent is capable of indicating one part in 20,000, the specimen must have contained in the outset 200 parts in 20,000, or one per cent. of albumen. This amount is rarely exceeded in pathological urine. Ordinarily it will be better probably for the physician to precipitate a specimen of the urine in a graduated test-tube by addition of potassium ferrocyanide solution and acetic acid, as recommended by Dr. Purdy,* estimating the quantity of albumen by the volume of the precipitate. A saturated solution of picric acid, used in large excess, is perhaps equally good.

The copper test which is generally relied upon for the detection of sugar is open to several serious objections. In the first place the reagent cannot be easily put up in a portable form. Test papers prepared with copper solutions soon deteriorate, as do most of the solutions (Fehling's, Pavy's etc.), themselves. The test pellets or compressed tablets have been found also to change, so that they cannot be relied upon. Parke, Davis & Co. put up a copper solution in little flasks hermetically sealed which keep well, and are useful for laboratory or office work, but cannot be conveniently carried in the pocket.

* See page 11 of this pamphlet.

On Bedside Urinary Tests.

BY GEO. OLIVER, M. D. LONDON, M. R. C. P. LONDON.

All busy practitioners must admit the clinical utility and importance of accurate, time-saving, and portable tests, by which they may, during their rounds, decide with precision and certainty, and on the spot, pathological conditions of the urine, or satisfy themselves, and their patients, if need be, without delay as to the soundness of that excretion. From the numerous contributions on portable urinary tests which have recently appeared in these pages, it is clear that practical men, who have long realized the serious inconvenience of carrying about caustic fluids for testing at the bedside,¹ are anxiously feeling their way to more manageable and handy yet equally trustworthy reagents, and I take it the profession at large is prepared to accept any useful suggestions towards this end. Hence the articles of Dr. Roberts on Acidulated Brine,² of Mr. Stephen on the Volumetric Estimation of Albumen,³ of Dr. G. Johnson on Picric Acid, and of Dr. Pavy. The corrosiveness of nitric acid and the causticity and instability of Fehling's solution, rather than want of faith in the trustworthiness of these tests, have doubtless mainly prompted the suggestion of more portable reagents. In the substitutes we therefore seek for compactness, handiness, and portability, without the sacrifice of the generally admitted reliability of the old forms for clinical purposes. Doubtless others besides myself have been trying to supply this desideratum, and may be prepared to offer useful hints. Having, however, attained to certain results which have satisfied my own clinical needs, I feel I should not delay further in communicating them to those of my professional brethren whom they may interest; and I do so with the hope that they may prove useful, especially in the saving of time to busy men, and may facilitate urinary investigation at the bedside. My experiments have embraced the qualitative and quantitative testing of albumen, sugar, and total acidity. I should make the preliminary statement that I have succeeded in all my reagents in abolishing the fluid state, and likewise the solid form, either of powder, crystal, or pellet. It occurred to me some time ago that evaporation of the test fluids I was then using on chemically inert filtering-paper, linen, or similar fabric, would secure the deposition of the reagents in a finely divided and concentrated state, a condition it was hoped favorable to such a rapid re-solution of them in the urine as to produce a quick and sensitive action on the constituents sought for. I soon discovered that my pieces of chemically charged paper were, when dropped into a small quantity of the urine in a test-tube, very delicate and cleanly tests; and being in the most portable and compact of all forms of clinical work, and, moreover, affording better results than I had previously obtained from the old corrosive test solutions, it was not long before I cleared my spoilt urinary case of the latter; and I can assure my readers I did so with a feeling of satisfaction and comfort. Then, inasmuch as it was an easy matter to graduate the papers with standard solutions of the reagents, I next proceeded to inquire how far this simple process could be carried in determining the quantities of albumen, sugar, and total acidity; in other words, I thus attempted to apply at the bedside the volumetric method of analysis in the form of pieces of filtering paper charged with definite quantities of the reagents with a quantitative color limitation on paper from which the percentage of the constituent sought for could be at once read off. Up to the present I am satisfied in having attained rapid and, for all practical purposes, sufficiently accurate results. I intend shortly, as a matter of curiosity, to scrutinize the figures indicated by my paper method for the estimation of albumen by the side of the burette with its standard solution; and I hope on some future occasion to be able to state definitely how near I can approach quickly at the bedside the results which can only proceed from the laboratory with its attendant delay. I must content myself in the present communication with my notes on—

THE QUALITATIVE ESTIMATION OF ALBUMEN.

When picric acid was brought forward by Dr. G. Johnson as a remarkably delicate test for albumen, it occurred to me as likely to assist in deciding the sensitiveness of the various albumen tests from clinical evidence rather than from the laboratory, to take a series of

¹ Hence the fact of many medical men having given up urine testing entirely during their rounds.

² *The Lancet*, vol. li., 1882, p. 613.

³ *Ibid.* vol. li., 1882, p. 324.

urines containing very small proportions of albumen, to subject them to all the best-known tests, and to carefully tabulate the results. Since then I have met with twenty urines, among others, which supplied the required condition, all being faintly impregnated with albumen presumably derived from the presence of a small quantity of pus, or of blood, or of both, as determined by the microscope.¹ The table of results annotated at the time of every testing is before me. All the urines were acid except one, which was alkaline. The reagents employed were the following:—

1. Strong nitric acid.
2. Boiling the sufficiently acid urine and afterwards adding dilute nitric acid.
3. Saturated solution of potassium ferrocyanide, and the urine freely acidulated by citric acid, as suggested by Dr. Pavy.²
4. Saturated solution of picric acid as advised by Dr. George Johnson.
5. Acidulated brine after Dr. Wm. Roberts.
6. Standard solution of potassio-mercuric iodide, after Tauret, and recently brought to notice by Mr. Stephen, with this modification—strongly acidifying the urine with citric acid instead of acetic.

The test fluid and the urine were in all the experiments brought into contact, as in Heller's method of using strong nitric acid, and the line of juncture was carefully examined for at least five minutes. Out of the twenty urines strong nitric acid failed to indicate the presence of albumen in sixteen instances, boiling in fourteen, acidulated brine in fourteen, and potassium ferrocyanide in twelve; while picric acid and potassio-mercuric iodide gave a distinct and generally a sharply defined ring of precipitated albumen in every case. The reaction was indicated by varying degrees of rapidity by the different tests; I must name the potassio-mercuric iodide and picric acid as the readiest; and of the two I would, if pressed for a preference, decide in favor of the former. I found as a rule strong nitric acid, acidulated brine, and potassium ferrocyanide much slower whilst bringing to light mere traces of albumen. I should here remark that I do not attach much clinical importance, so far as I can see at present,³ to the ability which the most sensitive of these tests possess in the detection of albumen in minimal proportions; for if I did not do so it might be justly thought by the practical physician that such observations as these indicate an ultra refinement in testing of no utility in daily work. They were simply made for the definite purpose of affording data suggestive of the most thorough of all the best tests for discovery of albumen in urine; and as such they undoubtedly point to the potassio-mercuric iodide and picric acid; and to potassium ferrocyanide and acidulated brine as next in order. I should remark that, with regard to potassium ferrocyanide, I am not quite satisfied that the method (Heller's) followed throughout these observations for the sake of uniformity in obtaining the comparative results, fairly put to the trial the capacity of this test as an albumen precipitant, for on several occasions I noticed the production of a very slight opacity all through the urine instead of a well defined ring. I am, therefore, with this qualification in my mind, inclined to think somewhat better of it than the above-recorded number of failures might lead anyone to suppose. The outcome of these observations, as well as more recent ones, suggests to me the grouping of the tests in the following rising order of power to detect small quantities of albumen: 1. Strong nitric acid and boiling. 2. Potassium ferrocyanide and acidulated brine. 3. Picric acid, potassio-mercuric iodide, and the two new tests brought forward in this paper. I have, as a rule, found the members of each group to be nearly equivalent, and confirmatory of each other; and, further, the albumen which strong nitric acid and boiling discovered was always detected with greater facility by all the other reagents, and those tests which comprise the third group frequently reveal traces which the others failed to bring to light; lastly, potassium ferrocyanide and acidulated brine certainly took precedence over strong nitric acid and boiling. As confirmatory of the foregoing observations I may mention that I lately supplied an analytical chemist with some strongly albuminous urine, and he subjected it in the following way to a comparative examination by strong nitric acid and the tests I am introducing in the paper form. After diluting the urine until the albumen was just detectable by the acid, he proceeded to further dilution, when the reaction failed to appear, though still the more delicate paper tests distinctly indicated the presence of the albumen. The albumen precipitants which I find work well as test papers are: potassio-mercuric iodide, potassium ferrocyanide, the two new tests, and picric acid.

¹ Whenever albumen was detected by any of the reagents mentioned in this paper, the urinary deposit was subjected to microscopical examination, and the impregnation was indicated, or at least suggested, by the presence of blood, pus, or casts.

² See *The Lancet*, vol. ii., 1882, p. 823.

³ Since writing the above, observations have, however, caused me to modify my first-formed impression as to the clinical value of the keenest albumen tests, and as at present known.

1. *Potassio-mercuric iodide* was introduced by Mr. Guy Neville Stephen to the readers of this journal¹ as an albumen precipitant discovered by Tauret of Troyes. I select it for production as a test paper because I formed a favorable opinion of it while working it side by side with the other tests, and because it enables one to readily determine the quantities of albumen at the bedside. I moreover found that this double halloid mercuric salt could be evaporated to dryness on filtering paper without impairing its albumen-precipitating powers; and the charge thus communicated still retains its integrity, though introduced over six weeks ago. Each paper contains one-fifth of a cubic centimetre of a standard solution, the formula for the preparation of which I hope to give in an article "On the Quantitative Estimation of Albumen at the Bedside," after my return from a holiday abroad. In the meantime I should say it differs considerably from that given in Mr. Stephen's paper. Hitherto, whenever the presence of albumen was indicated by this test paper, corroboration was furnished by picric acid, and the other tests of kindred power; and when the amount of albumen was such as to bring it within the range of less sensitive reagents, these also afforded confirmation. This mercuric salt is said to cause a quasi-albuminous precipitate in the urine of patients taking alkaloids; but I am not yet convinced of the correctness of this assertion, which I must leave, from want of conclusive evidence, neither positively confirmed nor denied. To this test I have repeatedly subjected the urine of one patient taking six grains of extract of opium every night and two grains of codeia twice a day, of another having two daily subcutaneous injections of morphia and atropia, and of another taking fifteen grains of salicine three times a day with this result: a very faint cloud on adding the paper and a delicate ring by the "contact" method of testing; but the same reactions followed the use of picric acid, and the microscope revealed the presence of pus cells. After giving up the salicine, the urine still gave the same faint indications of albumen in small quantity. But should this source of error exist, it is not likely to induce more than a semblance to the merest traces of albumen, and it is, moreover, easy to guard against it. Then, again, should a patient be taking an alkaloid, this series of test papers provides other equally sensitive albumen precipitants.

2. *Potassium Ferrocyanide*, when deposited to saturation on filtering paper, produces in conjunction with citric acid paper a reliable work-a-day test for the detection of albumen in urine. In my hands it has proved almost as sensitive as the other test papers here brought forward. The idea of ferrocyanide with citric acid originated with Dr. Pavy², who proposes to introduce them to the profession as a compound pellet. As thus presented, this albumen test will doubtless be compact, portable, cleanly and efficient. Perhaps, however, some practitioners may prefer to carry in their visiting lists, or otherwise, a leaflet of combined ferrocyanic and citric paper, rather than a bottle or other vehicle enclosing the pellets. The great convenience of this test paper, as of all the others, must appear, when it is known that at the bedside no further apparatus is required for the detection of albumen than a teaspoon or a wineglass,

3. *Two new albumen precipitants*.—During the past few weeks I have become acquainted with two precipitants of albumen in urine, which, so far as I know, have not received attention from the profession—and I am not aware that either of them has been applied as a urinary test. But until lately I felt some hesitation in introducing them to professional notice, lest by doing so I should unhappily create greater uncertainty as to the choice of the best and most generally useful albumen precipitant for clinical purposes than at present prevails; for I take it that medical men—at any rate many of the readers of the *Lancet*—are just now somewhat perplexed by the rival claims of picric acid, ferrocyanide of potassium, acidulated brine, and potassio-mercuric iodide on the one hand, and by their adhesion to the older methods, in which they have trusted so long, on the other. But favorable observations as to the clinical efficiency of these, what may be styled the latest competitors, and a desire to extend my paper method of analysis to other reagents, which may at least be sometimes usefully employed to determine results of a doubtful character, have decided me to ask my brother practitioners to give these new tests a trial by the side of the others.

(a) *Potassio-mercuric iodo-cyanide*.—While working with mercuric cyanide for another purpose, I found that when alone in saturated solution in water it failed to cause a precipitate in albuminous urine, highly acidulated by citric acid; but when mixed with potassium iodide, it threw down the albumen at once as a voluminous white cloud. The iodide and the cyanide combine, and produce a new double mercuric salt ($4KI, HgCy_2$), which crystallizes out of solution as beautiful colorless needles. When the solution of this iodo-cyanide is acidified by citric or other acid, a gas (hydrocyanic acid) is liberated, which when com-

¹ Op. cit.

² The *Lancet*, vol. ii, 1882, p. 823.

pletely expelled by heat, leaves the albumen-precipitating power of the solution unimpaired. It would therefore appear that this free hydrocyanic acid takes no part in throwing down the albumen, and that the precipitation must be ascribed to the mercuric iodide combination with potassium iodide. If so, this is but another form of the potassio-mercuric iodide test; but whether it will prove in some way more useful than the latter, I cannot as yet say. It can, however, be readily reduced to paper; and when thus employed, I have always found it as rapid and as sensitive as the other albumen test papers all of which have hitherto afforded corroboration of its integrity and trustworthiness as an albumen precipitant.

(b). *Sodium tungstate* is another delicate test for albumen in urine. According to the Journal of the Chemical Society for March, 1874, it is stated that this salt had been employed by F. L. Sonnenschein as a sensitive blood test, producing with ammonia a deep green color, even when the blood was so dilute as not to be recognizable by the spectro-scope, and as an albumen precipitant in the presence of acetic or phosphoric acid. I suppose this important observation has not attracted the notice of clinical observers, for I am not aware of any references to it in the medical journals in its obvious applications to urinary analysis. On mixing together equal parts of the saturated solutions of the tungstate (one in four) and of citric acid (ten in six), and of water, I obtained an albumen precipitant of great delicacy, rapid in operation, and one moreover, so far as I have ascertained, devoid of all objectionable qualities. When merely dropped into the urine, or used after the manner of Heller it has always quickly revealed the same minimal proportions of albumen as could only be brought to light by picric acid and by the other tests of equal keenness. But this combination, when evaporated to dryness on filtering paper, did not give results so satisfactory—they required about a minute to develop—as when, after previous acidification of the urine by citric acid, a paper charged with sodium tungstate only was used. The further capacity of this reagent as a detector of blood, at any rate in urine, I am sorry to say I cannot as yet confirm. Perhaps one or other of these albumen test papers will be found equally convenient in the consulting room as in the daily round; but should any practitioner prefer for the former a single solution, which will not stain the fingers, be free from all other objectionable qualities, and will, moreover, be stable and always ready for use, let me specially mention the acidified solution of tungstate, which has given me every satisfaction, and which I now prefer to any other liquid test for home use. Sodium tungstate is procurable as dry, non-deliquescent crystals, and is, moreover, very cheap; for a shilling will purchase as much as can be needed for the detection of albumen during the whole lifetime of the busiest practitioner.

Citric acid.—All the foregoing reagents are inoperative as albumen precipitants unless the urine is highly acidified; their application should therefore be preceded or accompanied by a sufficient charge of acid. For this purpose citric acid is easily made available when deposited to saturation on filtering paper, and in this form it has afforded me uniformly satisfactory results with all the albumen test papers.

Compound papers.—Instead of using citric paper separately prior to the reagent paper, it has been combined by a thin layer of rubber with the latter as a single test paper in the case of sodium tungstate and of potassio-mercuric iodide.

4. *Picric acid* can be deposited to saturation on filtering paper, which becomes a most compact and cleanly vehicle, and which, moreover, quickly delivers its charge to water. Repeated observation has shown me that when united with citric acid, as in the test papers, picric acid is divested of all the objections that have been urged against it. A few drops of albuminous urine instantly turns the bright picric solution extemporaneously prepared from the test paper, into a muddy one, while the addition of more urine does not redissolve the precipitate as when picric acid alone is used. For the detection of small quantities of albumen (less than 1 per cent.) the "contact" method of testing is necessary; then it is best to make the picric solution in a wineglass, to take it up by the medicine dropper, and to glide it gently over the urine in the test tube.

How to use the test papers.—About thirty minims of the urine are taken up by a nipple pipette, or medicine dropper, and transferred to a short test tube, preferably one about two inches in length. If turbid from urates, it should be gently heated. It is now strongly acidified by dropping into it a citric paper, which is shaken about for a few seconds, and may then be withdrawn or allowed to remain. It is not now necessary to ascertain if the urine is sufficiently acid, therefore without delay the test paper selected is allowed to fall into it. A simpler plan, and one which I find answers equally well, is to drop both the citric and the reagent papers into the urine, so that they may fall together to the bottom, and to one side of the test tube. The latter is now inclined, so that the urine may repeatedly and slowly flow over the paper; when, if albumen be present in small or medium quantity, a whitish cloud will very quickly gather above and below it, the more readily de-

tected by intercepting the light by the hand, etc.; while, in striking contrast, the upper part of the urine will remain clear. If, however, the albumen exists in large proportion, it will not usually produce a haze about the paper, but will coagulate on it, and will slowly fall from it in clots. Then, in any case, on shaking the tube the urine will become less or more opaque, according to the amount of albumen present. If, on the other hand, the urine preserves its brightness, or if any turbidity it possessed prior to the introduction of the test paper is not increased, it may be inferred it is free from albumen. But inasmuch as it is just possible, though I have never found it so, it may not have been sufficiently acidified to enable the reagent to throw down the albumen, it is advisable to dispose of this suspicion by adding another citric paper; when, if no precipitation occurs, albumen is absent. The whole proceeding, of course, takes up very much less time than that occupied in reading this description of it. The reaction is practically instantaneous when the urine has been freely acidified prior to the introduction of the test paper. It is, however, not so quickly obtained, though the delay only amounts to a few seconds, when, without previous acidification, the single compound test papers are used. The convenience and simplicity of the testing for albumen by these combined papers are very great, for the practitioner only requires to carry them in his visiting list or pocket book and to drop one into a little urine in a teaspoon or wineglass, when, after stirring it about for a second or two, the opacity of precipitated albumen will appear. Those who prefer to develop a zone of precipitation along the plane of contact of a test solution and the urine, can do so by aid of these papers. A test tube and a wineglass are required. Into the latter the reagent paper rolled up is placed with about fifteen minims of water, and, without shaking, is set aside, while a similar quantity of urine is put into the test tube with citric paper. After withdrawing the latter, the reagent, now in solution, is taken up by the pipette and is allowed to trickle down the side of the tube, in which it will collect at the bottom. After developing the ring, the two fluids may be shaken together, when the albumen will be more largely precipitated as a milky cloud.

The keeping power of the test papers.—None of the papers have been bottled or kept from the air and light during several weeks; on the other hand, they have been purposely exposed without covering. Still I cannot discover the least deterioration of their power to precipitate albumen or any change of color or of other physical quality. From the first I rather suspected the mercuric papers might not stand the exposure of daily work for any length of time; but I am now of opinion I over-estimated, if I did not misjudge, this possible source of failure. I am surprised they do not partake more than they appear to do of the deliquescence of the potassium iodide, which is merged in the new definite salts with which they are charged; for it was only after free exposure in a dampish room without a fire, a trial to which no delicate test should reasonably be submitted, that they became slightly limp, but even then without showing any diminution of their albumen-precipitating property. So far I can assert I have seen nothing to cause me to doubt the stability of even these reagents employed as test-papers, and they are the only members of this series of albumen precipitants which *a priori* might be thought at all susceptible to the deleterious effects of atmospheric influences. Though referring to a test touching another subject, I may say that my cupric papers, designed for the qualitative and quantitative estimation of sugar, and made over two months ago, are to-day as good and as sensitive as they were when freshly prepared.

In a leader of the *Medical Times and Gazette* for 1874¹ will be found the following passage, which tersely expresses the opinion of the writer as to the trustworthiness and adaptability to the requirements of medical work of the tests for the detection of albumen as then known: "The reader who has followed us so far will, we think, agree with us that none of the common tests are quite satisfactory, and that we still lack one which shall be cleanly, portable, cheap, and certain." I leave it for my readers to decide whether this remark still holds good.

In concluding these notes on the qualitative tests for albumen, I must state my consciousness of the imperfection of isolated observation, however faithfully recorded, and I would fain seek the scrutiny of many eyes, to correct if needful, or to more truthfully limit, any of the matters here advanced. With this object in view I have asked Mr. Hawksley, 357, Oxford street, W., to place a limited supply of these test papers at the disposal of the profession, and to furnish gratuitously the series to anyone who desires to put them to a careful trial, and who will kindly communicate the results of their observations to me or to *The Lancet*. In the meantime I hope that favorable evidence will not be long forthcoming to prove, as I have found, how satisfactorily they meet the requirements of daily work at the bedside, and to indicate which of them should be regarded as the best workable tests.

The Comparative Value of the Newer Tests for Albumin in Urine.

Although the mere presence of albumin in urine can no longer be regarded as anything further than presumptive evidence of damaged kidneys; yet, it has so long formed the basis upon which the diagnosis of renal pathology has been constructed, and, moreover, as an element considered with other symptoms it still holds, and is likely to hold, an important place at least in preliminary investigations of renal disease. Therefore, all matters concerning the most reliable and ready methods of detecting the presence of albumin in the urine must remain of practical interest and importance to the general practitioner. Much attention has been given of late to several newly proposed agents for detecting albumin in urine, both in this country and in Europe. Some of these, it has been claimed, are so exceedingly delicate in reaction as to detect the presence of albumin when the older methods, as by heat and nitric acid, failed to do so completely. Furthermore, most of these agents have been indorsed by some of the best authors on renal diseases, both here and abroad.

The general practitioner may, in consequence, encounter some confusion in deciding whether it be wise to accept the newer and readier methods or adhere to the older and long tried agents already in use. It is proper that we should carefully inquire into the relative value and reliability of the newer, as compared with the older tests, with the view of assigning to each their proper spheres in the routine of daily work.

First, then we shall note the acidulated brine test, brought forward by Dr. Roberts in the *Lancet*, Oct. 14, 1882. It is prepared by mixing together a fluid ounce of diluted hydrochloric acid with a pint of water saturating with chloride of sodium and filtering. It is claimed to be more delicate than Heller's test. It does not discolor the fingers or destroy the clothing in case it be spilled, and after a specimen of urine has been tested by brine for albumin, the same specimen may be tested for sugar by Fehling's test.

Picric acid was first brought forward by M. Gallipe in French current literature in 1872, Dr. G. Johnson more fully discussed its merits in the *Lancet* in Nov., 1882, as did also Dr. Pavy. Several articles appeared in that journal on the subject about the close of 1882 and the early part of 1883. The test solution is made by saturating boiling water with picric acid, and it may be applied by the contact method, viz., by allowing the picric acid solution to flow down a glass rod, or pipette, over the surface of the urine, which latter must be previously treated with saturated solution of citric acid. If albumin be present, a white cloud is perceptible at the junction of the two fluids.

Tauret's test, or the potassio-mercuric iodide solution, may be prepared as follows: potas. iodide 3.22 grammes, and hydrargyri bichloridi 1.35 gramme, are added to 100 cc. of distilled water and filtered. This salt was first proposed as a test for albumin by Tauret, of Troyes, in 1872. Mr. Guy Neville Stephen first called attention to its comparative value in the *London Lancet*. Its method of application is essentially the same as that of picric acid, previously having acidulated the urine with citric or acetic acid. It is claimed to be more sensitive in reaction with albumin than heat and nitric acid, that it is cleanly to handle, very permanent, and almost perfectly clear in solution.

Sodium tungstate was first proposed as a test for albumen by Dr. G. Oliver, of Harrogate, in the London Lancet, Feb. 3, 1883. His method of preparing the test solution is as follows: a mixture of equal parts is made of saturated solutions of sodium tungstate (one in four) and of citric acid (ten in six) in water. It is claimed to be "an albumin precipitant of great delicacy, devoid of objectionable qualities."

Dr. Oliver also proposes in the same number of the Lancet as a test for albumin, the potassio-mercuric iodo-cyanide, but as this is practically the same test as the one last described, it is needless to consider it further.

The potassium ferrocyanide test is simply a saturated solution of the above salt in water. Its application is made by previously acidifying the urine with acetic acid, and adding the ferrocyanide solution, when, if albumin be present, a white cloud is at once formed. This was proposed by Bödeker as a volumetric test for albumin. Dr. Pavy proposed subsequently to make the test with citric, instead of acetic acid, and he also introduced the pellets, (compressed) of sodic ferrocyanide and citric acid, which are simply crushed and placed in a test tube containing the suspected urine, agitating without heat.

Dr. Oliver first prepared all of these tests, save brine, in portable form, so that they could be carried in the pocket-book, and applied at the bedside of the patient. His method consisted in saturating chemically inert filtering paper with the test solutions, and cutting the papers into small strips. An acid paper was also prepared by saturating filtering paper with strong solution of citric acid. The application of these tests is as follows: A small test tube is used, with a capacity of say 60 minims. This is filled about half full of the suspected urine, and a citric paper is rolled up and thrown into the tube. After agitating for a minute or so one of the other papers is added, say the potassio-mercuric iodide, or ferrocyanic, and if albumin be present, a white cloud at once forms about the last paper introduced.

Dr. Oliver has also prepared what he terms the compound papers, made by joining together a citric paper with one of the others. This simplifies the application of the test, inasmuch as but one paper need be used in testing. A full account of these test papers and their application is best told in Dr. Oliver's own words,* which is done in the London Lancet, Jan. 27 and Feb. 3, 1883, or in a small book issued by himself on "Bed-side Urinary Tests."

The carbolic acid test, in any of its forms, I have not used, for reasons which will become obvious to any one who tries it in detecting the smaller traces of albumin.

The above, then, in the main, are the newer tests proposed for detecting albumin in the urine. It is proper that we next inquire into their advantages and disadvantages, in order to properly estimate their relative values. What I shall say upon this point is the result of a series of experiments with all of these agents, extending over the past eight months, or perhaps a little longer.

As to the test papers, Messrs. E. H. Sargent & Co., 125 State street, Chicago, kindly prepared, at my suggestion, about ten months ago, all these papers, according to Dr. Oliver's method; and as they have been in stock since, they may be procured by any one desiring to try for themselves. I also procured, about eight months since, through Mr. Hawksley, 357 Oxford street, London, two complete series of Dr. Oliver's own papers, for the purpose of trial; and with all these, I have made careful and repeated experiments. I prefer the potassio-mercuric iodide and tungstate papers to all others.

The most serious objection, perhaps, to all these newer tests, is the fact that they give a reaction with other matters, occasionally present in the urine, such as peptone, mucin, parapeptone, and urates, and, moreover, if the patient be taking certain of the vegetable alkaloids, (notably quinine), or oleo-resins, or some of the alkalies, or even alcohol in considerable quantities, before the urine is voided, a similar reaction occurs, none of which are distinguishable from albumin precipitate, without the additional employment of heat to the test; and the latter in some cases decomposes the test solution, (notably the ferrocyanic).

I think it may be laid down as a rule, to which I at least know of no exception, that no test for albumin in urine is absolutely reliable unless combined with or followed by heat. At a temperature of 72° to 75° C. serum albumin coagulates and becomes insoluble in acid solutions; and if of any quantity, the insolubility is perceptible in clear solutions in the formation of a more or less opaque cloud. I know of no other bodies met with in the urine, if that fluid be clear, and acid in reaction, which will give a similar reaction.

*See page 4 of this pamphlet.

Of course, in dealing with urine where the amount of urine is large, most of the ordinary tests are sufficiently accurate to determine its presence beyond reasonable doubt; because phosphates, urates, peptones, mucin, bacteria, or other bodies, which may give a precipitate simulating closely albumin, are almost never present in sufficiently large quantities to throw down a coagulum of the density, appearance and extent of albumin when present even in moderately large quantities. And hence in those precipitates, filling, say a third, or half of the test tube, we are reasonably safe in assuming at least that albumin is present, whatever else may be also, though the reaction be brought about by most any of the tubes in ordinary use.

As to the brine test of Dr. Roberts; it is cleanly, does not soil the hands, or clothing, keeps well, is easily applied, and is a very delicate reagent. In testing for very small traces of albumin, however, it is inferior to two or three others, and one reason for this may be, that it requires so large a quantity of the solution to cause a reaction, that it very much dilutes the urine, rendering detection of albumin in small quantity more difficult.

The sodium tungstate test does not stain, is very clear (the most so of all); its reaction is a little slower than some of the others, but extremely delicate, and properly applied is one of the best of all. It will not answer to apply this test as some of the others have done, namely, to acidify the urine first, and then add the tungstate solution, because an excess of the tungstate may get into the urine and render it inoperative, for it must not be forgotten that tungstate of sodium solution in excess in the presence of an acid dissolves both egg and serum albumin. We must thus use the test solution, as advised by Dr. Oliver,* already described, to make it reliable.†

The picric acid test seems to have excited more discussion than any of the others. It will quickly react with albumin in small quantities, but it has some serious drawbacks for general use. If allowed to mix with the urine (and it is not always possible to prevent this) it colors that fluid yellow, which is unfavorable for detecting faint traces of albumin. Again, if picric acid comes in contact with the fingers (and it often does in manipulating) it leaves ugly stains behind for many days.

In a paper on "Clinical Diagnosis of Bright's Disease," which I presented to the Chicago Medical Society a year ago, I mentioned that I had been in the habit for years of estimating the approximate quantity in the urine by the potassio-ferrocyanide solution and acetic acid. Subsequent use in my hands shows that it much less frequently produces a precipitate with other bodies than albumin, in comparison to the other newer tests.

My reasons for using the test so much in daily work are as follows: In addition to being a fairly sensitive test, as to presence of albumin, it is a fair working volumetric test. I do not mean by this that it is to be absolutely relied upon as an accurate volumetric test, but rather that it will indicate sufficiently accurately the relative amount of albumin from day to day to make it valuable as a ready method of comparison in following cases of albuminuria. In using the test, I use a graduated tube, and as the albumin subsides to the bottom after a few hours (twelve) its quantity by bulk may be read off from the scale on the side of the tube and noted at each examination. I now do not use this solution in detecting the presence of albumin, because, in doubtful cases, the test can not be corrected by heat; the latter at once decomposes the ferrocyanide salts with an acid, liberating hydrocyanic acid, and producing a white turbidity in the solution, which might be taken for albumin.

The potassio-mercuric iodide solution is an extremely delicate test (about equal to tungstate), very quick in reaction, almost clear (slightly bluish), keeps well and is readily applied.

I have noted one objection which the tungstate does not possess, namely, after the test is applied, it cannot be set aside and examined again satisfactorily. This potassio-mercuric salt, in the presence of an acid, soon begins to decompose, liberating free iodine which discolors the solution and renders faint traces of albumin difficult to distinguish. This does not so materially detract from the value of the test, as the reaction is not immediate, and it is slow.

Now as to the relative sensitiveness of these tests. Dr. Oliver says, "as a result of many observations and experiments, that in his opinion, one part of albumin may be discovered in 20,000 by the iodo-mercuric, picric and tungstate tests; in 10,000 to 12,000

* See page 7 of this pamphlet.

† I have observed that by previously heating the albumin solution to 212° the tungstate in excess will not dissolve it afterwards.

by the ferrocyanide and brine tests, and in 6,000 to 7,000 by the heat and nitric acid tests."

The following are the details of some of my experiments, made with the view of satisfying myself on this point:

The urine of one of my patients who was convalescing from acute Bright's disease, was subjected to experiment. The urine, when experimented upon, contained a trace of albumin, was acid in reaction, sp. gr. at 60° F. was 1022, and contained a few epithelial casts.

1. On the undiluted urine all tests showed the presence of albumin.

2. 50 cc of the urine was diluted with 50 cc of distilled water, and the following noted: Reaction still apparent with all tests.

3. 50 cc of the above was diluted with 50 cc of distilled water, and the following noted: Reaction perceptible with mercuric, tungstate and picric acid tests. The others were negative.

4. 50 cc of the above was diluted with 50 cc of distilled water, and the following noted: Mercuric and tungstate tests showed faint change; all the others were negative.

The urine of a patient under my charge, who was suffering from slight chronic nephritis (parenchymatous) was next subjected to experiment. The urine, when experimented upon, was normal in appearance and reaction at 60° F., sp. gr. was 1025. A slight trace of albumin was present. The microscope showed a few granular casts and renal epithelium.

1. On the undiluted urine all the tests gave reaction, showing presence of albumin.

2. On diluting 50 cc of the urine with 50 cc of distilled water, the following was noted: Mercuric, picric and tungstate tests gave reaction. The other tests were negative.

3. 50 cc of the above was diluted with 50 cc of distilled water, and the following noted: The mercuric and tungstate tests still gave faint turbidity. The other tests were negative.

4. 50 cc of above was diluted with 50 cc of distilled water, when all the tests failed to show any change whatever.

The urine of a patient who was suffering from chronic interstitial nephritis (granular atrophy) was subjected to experiment. Having been under my charge for a year and a half, I knew his urine to average a small amount of albumin (6 to 8 per cent. by bulk) and to contain a few hyaline casts, sp. gr. (at 60° F.) ranged from 1008 to 1018, and reaction usually neutral or alkaline.* It was the latter when experimented upon.

1. Observations on the undiluted urine showed presence of albumin by all tests.

2. 50 cc of the urine was diluted with 50 cc of distilled water, when all tests still showed presence of albumin.

3. 50 cc of above was diluted with 100 cc of distilled water, and the following was noted: Mercuric, tungstate and picric tests showed faint reaction. All others negative.

4. 50 cc of above was diluted with 50 cc of distilled water, and all tests failed to show any perceptible change.

These experiments, and in fact all my experiments, were conducted with the same quantity of urine or solution operated upon in each case, and similar size and make of test tubes were used in all cases. Now with reference to the reaction of certain of these newer tests in dilutions beyond the point where heat and nitric acid fails to give any change. Is this reaction albuminous? If it be albumin in minute traces, which these so-called more delicate tests show, where heat and nitric acid fail; it would go far towards proving that Gubler first suggested, in 1865, and afterwards Ultzman, in 1870; and J. Vogel, in 1873; and Johnson, of London, in 1883: namely that traces of albumin may be a constituent of normal urine. While I know of no reason why traces of albumin (probably untransformed) should not be present in normal urine (as sugar is now known to be), if, as M. Leube† claims repeatedly, to have found it present in cutaneous sweat. But before accepting the above as fact, it must first be shown conclusively that certain substances known to exist in urine, closely allied to albumin, are not the ones which these newer tests give reaction with instead of albumin. Bence Jones refers to a peculiar case of this kind.‡ Baylon describes an albuminoid substance under the name of

* Autopsy since showed both kidneys contracted and granular; the left weighed 2¼, and the right one less than 3 ounces.

† Virchow's Archiv, Band 48, p. 181.

‡ Annalen A. Chem. u. Pharm., Band 67, pp. 97 to 105.

albuminose, which is said to occur also in normal urine. Peptone-like bodies were found by O. Schultzen and L. Reiss in urine after phosphorus poisoning.* The identity of this peptone-like substance with true albumin peptones is still doubtful. Finally, according to Bechamp, a protean substance can be precipitated from every normal urine by three times its weight of 88 to 90 per cent. alcohol, which, after washing, is soluble in water. Bechamp has given it the name of nephrozymose.† It has furthermore been stated on high authority, that no test yet known will detect albumin in traces more minute than will the old heat and nitric acid test.

I have conducted a number of experiments with the view of throwing, if possible, more light upon this question, from which I select the following as examples:

First, I prepared a sample of pure blood serum from a healthy man, secured during traumatic hæmorrhage (epistaxis).

1. 50 cc of the above blood serum was diluted with 100 cc of distilled water and double filtered, and on testing the following was noted. The mercuric, brine, tungstate, ferrocyanic and heat and acid (nitric) gave very distinct reaction, showing albumin.

2. 50 cc of the above was diluted with 100 cc of distilled water, and still all tests gave reaction (positive).

3. 50 cc of the last dilution was mixed with 100 cc of distilled water, and the following was noted. The mercuric, tungstate and brine tests gave the most distinct change. Heat and nitric acid, and ferrocyanic tests were still positive, but considerably fainter.

4. 50 cc of last dilution was added to 50 cc of distilled water, and the following noted. Mercuric and tungstate tests gave the most distinct change. Heat and nitric acid and brine were the faintest.

5. 50 cc of last dilution was added to 50 cc of distilled water, and the following noted. Heat and nitric acid practically failed to show any change, as did also brine. The mercuric and tungstate tests showed distinct change (positive).

6. 50 cc of last solution was still further diluted with 50 cc of distilled water, and the following noted. Heat and nitric acid and brine failed (negative). The mercuric and tungstate gave distinct change (positive). The temperature of the mercuric and tungstate tests was raised to 212° F. over a spirit lamp. The opacity was not in the least cleared thereby.

7. 50 cc of last dilution was further diluted with 50 cc of distilled water, and the following noted. The mercuric and tungstate tests gave perceptible reaction (positive). The others failed (negative).

8. 50 cc of last dilution was still further diluted with 50 cc of distilled water, and tests applied as before. The mercuric and tungstate tests both showed very faint but perceptible change (positive). The others all failed as before.

9. 50 cc of the last dilution was further diluted with 50 cc of distilled water. The mercuric and tungstate tests now failed to show any change whatever. A tube of the diluted serum was compared with the tungstate and mercuric tests, and examined in various lights, but no difference could be distinguished.

The next experiment in this connection was as follows in general terms: The serum was obtained from a vesicated surface on a healthy man (save neuralgia). The agent employed to produce the blister was cathartical collodion. The experiments with this serum were conducted precisely similar to the last described, and gave substantially the same results.

I next experimented upon the fresh blood serum (18 hours after death) of a healthy pig; the blood being withdrawn from the carotid artery in killing the animal for use. The experiments with this serum were conducted similarly to those on the human blood serum, and the results were almost the same. A slight difference in favor of the heat and nitric acid test seemed to exist, that is to say, the heat and nitric acid test seemed to give positive reaction a little longer than in the case of human serum, but the mercuric and tungstate tests unquestionably detected the albumin diluted beyond the point where heat and nitric acid failed.

I next took 10 cc of human blood serum (from an epistaxis case,) and after diluting it with 90 cc of distilled water, I precipitated the paraglobulin therefrom, and double filtered. The solution was submitted to the same tests with substantially the same results, the mercuric and tungstate tests unquestionably showing distinct reactions consid-

* *Annalen des Charité Krankenhauses zu Berlin*, Band 15, p. 9.

† *Neubauer and Vogel's Analysis of Urine*, p. 100, (1879).

erably beyond the point of dilution, where heat and nitric acid failed to show any change.

In the case of egg albumen, however, the results will be found altogether different, as I have observed repeatedly; for instance, I took a weak solution of egg albumen in distilled water, and selecting as the most delicate tests, the potassio-mercuric iodide and tungstate of sodium, I compared them carefully with the results of the heat and nitric acid tests, which were as follows:

1. On the solution, as prepared, all three tests reacted distinctly (positive), showing albumin precipitate.

2. 50 cc of the solution was diluted with 50 cc of distilled water, when all three tests still showed reaction positive.

3. 50 cc of last solution was next diluted with 100 cc of distilled water, when all three gave reaction, though faint. The heat and acid, if anything, were most distinct.

4. 50 cc of last solution was next diluted with 50 cc of distilled water, and the following noted. The heat and acid and mercuric tests gave a faint but perceptible change, distinguishable only on comparing them with a similar sized tube of distilled water, in front of a dark background. No change could be perceived with the tungstic test.

5. 50 cc of last solution was next diluted with 50 cc of distilled water, and the three tests applied. On holding them side by side, in various lights, no appreciable difference could be distinguished between any of the tests and a similar tube of distilled water, though examined by several individuals.

Thus it would seem there is a greater difference between the two native albumens (egg and serum) than has been generally supposed. I believe that heat and nitric acid is a peculiarly delicate and appropriate test for the former, as I have noted in testing, in the presence of minute traces of egg albumen in distilled water, that the reaction produced by heat and nitric acid is considerably more pronounced than by any other test. The coagulum seems more dense and opaque, but it is about equal to the mercuric test as to minuteness of quantity it will detect.

Now as to physiological albuminuria, so-called. It is surprising how many people in apparently perfect health have small traces of albumin in their urine; even by the heat and nitric acid test, if properly and carefully applied, it will probably show traces of albumin in 8 or 10 per cent. of such individuals. I believe, as a result of my own observations, that the majority of people of all ages have traces of albumin in their urine, either occasionally or constantly, as determined by the mercuric and tungstic tests.

A number of observers, Senator, Leube, and lastly Chateaubourg, have recorded results from examinations on a large scale, which are quite startling to believers in the pathological significance of albumin in small quantities in urine. Chateaubourg, of Paris,* within the past year, has made a large number of examinations on healthy individuals, of which the following are a few of the results: 98 soldiers, from twenty-one to twenty-five years of age, were first selected and their urine obtained on Monday (hence there was no fatigue duty the day previous), three hours after the midday meal. Of the 98 samples, 44 showed the presence of albumin.

In July, 1883, he tested the urine of 94, five hours after a meal, and 76 contained albumin, 8.2 per cent. He examined, at the Hôpital des Enfants Assistés, the urine of 142 healthy children, from 6 to 15 years of age, 111 of which contained albumin. Of 231 soldiers, on fatigue duty, whose urine was examined, he found albumin in 201 cases. I believe the test used by Chateaubourg was the potassio-mercuric iodide. It cannot be claimed, that in all these cases the albumin was the result of pathological change; however slight; for Chateaubourg distinctly says, the urine of many of these cases was submitted to the microscope without finding any evidence of kidney lesion.

I have personally obtained very similar results from observations on a considerable (though smaller) number of cases. I have examined the urine of many healthy persons, where the urine was otherwise normal, save that it showed the presence of albumin on the application of the mercuric and tungstate tests, but no reaction could be found with the heat and nitric acid, however applied. When I say, the urine of these persons was normal, I mean in appearance, *sp. gr.*, in quantity and reaction, and I carefully examined it with the microscope, going over several slides, but failed to find casts, renal epithelium, or bacteria.

* Chateaubourg *Recherches sur l'Albuminurie Physiologique*, Paris, 1883.

It may be thought, perhaps, by strong believers in the delicacy of the heat and nitric acid test, that where it failed, and other tests showed albuminous reaction, the failure may have been due to want of the precautions necessary to insure success. So far as my own manipulations at least are concerned, this could not very well be the case. I am aware of the fact, that of all tests the heat and nitric acid requires the most care and precaution to insure accuracy.

One of the most serious objections to the heat and nitric acid test, especially in cases where albumin is present only in small quantity, is the fact that if the acid be not added in sufficient quantity, we sometimes fail to get any change; and, on the other hand, if too much acid be added, it will dissolve small traces of albumin; thus in either case the test may be negative.

My friend, Prof. Walter S. Haines, of Rush College, has suggested to me a method of procedure, which I think obviates this difficulty. In applying the nitric acid to the urine, he inclines the test tube quite obliquely, and thus allows the acid to flow slowly down the side of the tube and through the urine (which latter it does owing to the greater gravity) to the bottom, leaving behind, the urine acidified in increasing intensity, each layer from the surface to the bottom. Some of these strata will be found of exactly the proper acidity to give the albuminous reaction.

In all cases where heat and nitric acid failed, and other tests showed any change, the former was given the benefit of most careful application, and repetition in several forms, among others the double method of Dr. Brown-Sequard* was always tried; also the method of complete cooling and re-application of heat as advised by Tyson.† In fact, the heat and nitric acid test was given by far the most care and attention of all, because, after the results obtained with it on egg albumen, I was working under the conviction that the heat and acid test would, if properly applied, reveal the presence of albumin, where any other known test would do so, for I had made my observations on egg albumen before I had experimented upon blood serum.

From the sum of my observations I draw the following deductions:

First:—That certain of these newer tests, as the potassio-mercuric iodide, sodium tungstate, picric acid, and perhaps the brine and ferrocyanic, will detect serum albumin in more minute quantities than will the heat and nitric acid test.

Second:—That the most delicate and reliable of these, and possessing the fewest objections, are the potassio-mercuric iodide and sodium tungstate tests.

Third:—That the test papers of Dr. Oliver, especially the sodium tungstate and potassio-mercuric iodide, are handy measures for preliminary examination of urine at the bedside of the patient for determining the presence of albumin in urine,

Fourth:—That to be entirely reliable, the correcting influence of heat must be employed in applying all these newer tests.

Fifth:—That the potassio-mercuric iodide, and sodium tungstate tests, and also the test papers of Dr. Oliver, are undoubtedly valuable acquisitions to our resources; inasmuch as through their greater delicacy and more ready applicability, they are likely to lead more frequently to resort to the microscope, and thus detect the early stages of certain forms of nephritis, which might otherwise escape the observation, till too late to save or prolong life.

Sixth:—That the question of the near future, as to albumin in urine is likely to be, not only is it present, but what quantity of albumin in the urine constitutes a pathological condition? and this question must be largely determined by the microscope.

*Archives of Scientific and Practical Medicine, 1873.

†Practical Examination of Urine, 1883.

Urinary Test Papers.*

BY C. W. PURDY, M. D., FIRST VICE-PRESIDENT CHICAGO MEDICAL SOCIETY,
MEMBER STATE MICROSCOPICAL SOCIETY OF ILLINOIS.

Mr. President:—I need scarcely remind you, that anything that can throw additional light on the pathological conditions of the urine, and render more ready the detection of these changes, is of the very deepest importance to the medical profession at the present time. Hence the subject of urinalysis is assuming greater importance day by day.

There is scarcely a well-regulated hospital now in our land, which does not insist on the rule of a careful examination of the urine of every patient on admission, and experience has already amply confirmed the wisdom of such a measure.

Already the leading life insurance companies have discovered, that unless the same rule is insisted upon in the case of applicants, many hazardous risks are overlooked by the ablest diagnosticians which they are able to command; and five years hence, no company will dare incur the risk of neglecting this precaution.

Furthermore, the day is already dawning, when no reputable surgeon can afford to employ the knife, or chloroform, without previous observation of the same precaution.

The important, and often quite unexpected light, which a casual examination of urine sometimes throws upon a case, has become of such repeated and almost daily experience, that it can assuredly no longer remain a question, but it is the plain duty of the conscientious physician to analyze the urine of every patient whom he is called upon to attend.

I have repeatedly alluded elsewhere to the fact, that the most hopeless of all forms of nephritis begins its march so silently, and stealthily, and proceeds to quite an advanced stage, indeed often to the very verge of extinction of life, with attendant symptoms so obscure that the very ablest diagnosticians repeatedly overlook the presence of this grave malady, unless they make it a rule always to examine the urine of patients. I refer to interstitial nephritis (chronic Bright's disease). Not alone is it important to make the early discovery of these grave renal lesions, when they are yet within range of medication, but also, those quasi-pathological states of urine, which are likely to lead to damaged kidneys, are important points, which the improved methods of urine-testing, recently brought forward, enable us readily to detect.

The means hitherto at our command, in searching for both the presence of albumin and sugar in urine, have possessed so many disadvantages and inconveniences, necessitating the expenditure of so much of the valuable time of the busy practitioner, that any measures which will shorten the method, and expedite the process, are sure to prove of the greatest value to the profession.

Something over a year's practical experience with test papers, as first suggested by Dr. Oliver, has convinced me that papers could be brought to such a condition of perfection, as practically to revolutionize the old system of urine-testing, by the cumbersome and corrosive chemicals hitherto employed. Moreover, they possess the great advantages of portability, and ready applicability at the bedside of the patient.

In the Journal of the American Medical Association, Jan. 19, 1884, I called attention to the comparative value of the newer tests, and described the preparation and history of the albumin precipitants employed, both here and in Europe, to date.† It is my purpose now, to supplement this with a description of the application of both the albumin and sugar tests, in paper form, and to point out some of the advantages they possess over the old methods.

Messrs. Parke, Davis & Co., of Detroit, have kindly prepared at my suggestion,

*A paper read before the Chicago Medical Society, April 14, 1884.

†See page 9 of this pamphlet.

the series of albumin precipitants in paper form, and moreover, they are entitled to the credit of carrying into practical use, the idea of arranging also with the same series, those for sugar, in such a beautiful, compact, and portable form, as to leave very little to be desired. The profession is certainly under obligations to this enterprising firm for this extremely neat and practical work, a sample of which I have the pleasure to show you this evening. First then, we shall consider the papers used in qualitative testing for albumin. Experience has demonstrated that the four solutions, ferrocyanic, picric, iodo-mercuric and tungstic, produce the most efficient tests in paper form. All of these are very soluble salts, thus rendering it an easy matter to heavily charge the papers with these solutions, which in turn are readily discharged in the urine under examination, and the stability of the papers is such that many months exposure to the atmosphere does not impair their powers, which remain quite equal to the solutions from which they are made.

It has been urged as an objection to the use of these tests, that they throw down other bodies than albumin, occasionally met with in the urine, unless so many precautions are observed, that it is apt to confuse the amateur in his manipulations. Experience will demonstrate, on the contrary, that in searching for albumin with these tests, they possess the elements of rapidity, simplicity, and accuracy, to a degree not approached by the nitric acid, or any other method at present known. The precipitates produced by any, and all of these papers, as Dr. Oliver properly observes, "In ninety-nine cases in a hundred, are albuminous," and if the one precaution alone of heating afterwards is observed, the precipitate remaining is positively albuminous, in all cases. Surely nothing can be more simple and easy then, for detecting albumin in urine.

But the very objections which have been raised to the use of these tests, namely, their liability to precipitate various bodies occasionally present in the urine, if these reactions are but properly comprehended, render them valuable above all other methods, where we wish to extend our knowledge of the condition of the urine, beyond the mere presence of albumin and sugar, and such knowledge is every way desirable, if we seek to know the manner in which the kidneys are discharging their function.

Aside then from the mere presence of albumin in urine, which can be so readily disposed of, let us next ascertain what additional information these tests are able to give us, bearing on the physiology or pathology of the urine.

The mercuric iodide test first claims our attention as the most delicate of all, as an albumin precipitant. This test, in addition, is a most sensitive one for alkaloids; and hence if the patient be taking these, such as quinine, it throws them out of solution in the urine, producing a haze, at first sight much resembling albumin. You will observe, however, that in the case of the alkaloids, the haze is a uniform turbidity in the solution, which does not break up into flocculi, or settle to the bottom, as in the case of albumin. Gentle heat will of course clear up the turbidity from the alkaloid. It is not necessary to use the citric paper first, to detect alkaloids; they are thrown down at once by the mercuric test. Oleo-resins, as balsam copaiba, are not thrown down by mercuric iodide in the absence of an acid, but it precipitates peptones, if present in the urine, re-dissolved by heat.

Picric acid papers throw down alkaloids, peptones, and occasionally urates, if present in the urine, the latter in excess. The picric, unlike the mercuric test, throws down oleo-resins, even in the absence of an acid, as a dense opacity. This dissolves on boiling, and reappears on cooling.

The tungstic papers differ from the picric and mercuric, in the important particular that they will not cause precipitates with alkaloids; but like the two latter, in the absence of an acid, they will not precipitate oleo-resins.

The ferrocyanic papers are likely to prove the most valuable of the series, being the least liable to error. So far as at present known, they throw down precipitates with no bodies usually met with in the urine, save albumin and urates. The latter may be avoided by first diluting the urine with equal bulk of water. This, then, is the only known test which will detect serum albumin in the urine, positively, without heat.

I had long ago observed, and in fact stated in the American Medical Association Journal, Jan. 19th, 1884, that "Its use in my hands, shows that it much less frequently produces precipitates with other bodies, than albumin, in comparison to the other new tests." I had not discovered, however, till afterwards pointed out by Dr. Oliver, that urates were the only bodies precipitated by this test, save albumin, and the importance of this point is very significant; for, as Dr. Oliver says, "In searching for albumin with this test we have only to be on our guard against urates as the only possible source of error," and the means of avoiding this has been referred to above, even without the employment of heat.

The citric papers accompanying these tests, are destined to cover a wider range of

utility, than the mere previous rendering of the urine acid, and clearing up phosphatic turbidity. In a recent interesting personal letter, Dr. Oliver has called my attention to the fact, that if the citric paper be first used, before one of the albumin precipitating-papers is added it eliminates two or three of the possible errors, (a) mucus, if present, is immediately precipitated by the citric paper alone, causing a milky turbidity; (b) oleo-resins, likewise are thrown out of solution, forming an opacity; (c) excess of urates, detected readily by precipitation with citric paper.

The test papers also open up another field for study, as they not only detect the presence, but also differentiate the form of albumin in urine. It is well-known that albumin may be met with in the urine in three forms: ordinarily as serum albumin; or it may be associated with an acid, when it is known as acid albumin; or, it may be associated with an alkali, when it is termed alkaline albumin. These papers, like heat, precipitate all modifications of albumin. Dr. Oliver says, "the observer, if he wish to do so, may differentiate serum albumin by heat, when the solvent power of the acid paper may be called in. Alkaline albumin may likewise be sought for by heat, aided by the previous use of the citric paper; and acid albumin may also be precipitated by it (heat) after neutralizing, (care being taken not to alkalinize) the acid, by means of the carbonate of soda paper. The separate use of these papers, therefore, enables the observer to utilize heat, not merely as a detector of serum albumin, but also as a discriminator of the three modifications of albumin."

There are two weighty advantages possessed by these new tests over the older methods, which should command their general adoption. Their surpassing delicacy, and ready applicability at the bedside, alone entitles them to precedence. All of them will unquestionably discover minuter traces of albumin than will the nitric acid method. This may readily be proven, by taking a given specimen of albuminous urine, precipitating first all of the albumin it is possible to do with nitric acid and heat; then if the precipitate be filtered off, the filtrate may be shown to contain a trace of albumin, by most any of these tests (especially the mercuric and tungstic).

There are cases, such, for instance, as slight or intermittent albuminuria, sometimes accompanying gout, or preceding intestinal nephritis, where these tests afford us a more certain means of detecting the earliest manifestations of danger, which may often be postponed or avoided. Again, in acute febrile diseases, accompanied by albuminuria, which frequently lay the foundation for permanent damage of the kidneys, such as diphtheria, erysipelas, and, above all, scarlatina, it is clearly the duty of the physician to follow such cases to the complete disappearance of all traces of albumin from the urine, as the only safety of the patient from future danger. With such agents as the mercuric and tungstic tests, the slightest trace, even to one part of albumin to twenty thousand, may be detected, and hence there can no longer be any excuse for the physician who assumes the responsibility of discharging the scarlet fever patient as cured, to have such cases return in a few weeks with dropsy, and, perhaps, permanently damaged kidneys.

MODE OF USING PAPER TESTS.

Urine if turbid, should not, of course, be submitted to any test, if we expect to gain accurate information. If not clear when passed, it should be filtered; if this does not clear it, a fourth of its bulk of liq. potassæ may be added, then warmed and filtered; if still not perfectly clear, one or two drops of the magnesian fluid (magnes. sulph., ammon. chlorid., aa 1 part, aqua dest. 8 parts, liq. ammon. pur. 1 part) may be added, then warmed again and filtered.

Thirty to fifty minims of the suspected urine may be poured into a sixty-minim test tube, and its reaction first noted with litmus paper. If slightly acid, one citric paper is introduced, but if alkaline, two or more must be used to insure acidity. Next, one of the albumin precipitants is added and any change noted. Instead of above method, both the citric paper and the albumin precipitant may be allowed to fall into the test tube together. If albumin be present, a milky cloud will quickly form about the papers, changing slowly into flocculi, and shortly settling to the bottom of the tube. Such is the usual reaction with small quantities of albumin, say below one-sixth of one per cent. But if albumin be present in larger quantity, it will not produce a generally diffused milkiness through the solution, unless the tube be shaken, but will coagulate about the papers in solid lumps, and fall in pieces at the bottom. Other methods are described by Dr. Oliver, which may be resorted to if desired. He says, "Those who prefer to develop a zone of precipitation along the plane of contact of a test solution, and the urine, can do so with these papers as follows:

Two tubes, or a tube and a wine-glass are required. Into one the reagent paper, rolled up, is placed, with about fifteen minims of water, and set aside; while a similar quantity of urine is put into the other tube, with a citric paper. The reagent (now in solution) is taken up by the pipette and allowed to trickle down the side of the tube in which it will either glide over the urine, or collect below it." I think it will be found preferable not to agitate the contents of the tube in testing; for it will be observed that all the urine above the papers will remain clear, offering a strong contrast to the milky precipitate of albumin below. In this connection I may mention a suggestion by Dr. S. C. Smith, of Halifax, which is to bend the papers into a circle, so as to fit inside the test tube, and then push them down, say, within an inch of the bottom. The tube is then filled with urine. If albumin be present, the whole of the urine below the papers becomes opaque, while that above them remains transparent and unchanged.

TEST PAPERS FOR SUGAR.

All agents employed in testing for sugar, are either caustic, alkalies, or bodies associated with an alkali. The principle of construction of test papers for sugar depends upon the fact that in the latter case it is not always necessary to use a caustic alkali. For instance, in the cases of mercuric cyanide, picric acid, and Trommer's copper test, the substitution of soda carbonate for the caustic alkali will answer quite well. Papers, therefore, may be made charged with such agents, and separate papers, charged with soda carbonate, may also be kept in the case for use; or, the paper in some instances may be doubly charged with the reagent and the soda carbonate together; thus necessitating the use of but one paper in testing for sugar. The first I think will be found the better way in most cases, for two reasons: First, we may meet with a saccharine urine of exceptional acidity, rendering it necessary to add an extra paper of soda carbonate. This contingency may be met in another way, by being careful to operate upon only a small quantity of urine at a time. The second and most important advantage of separate papers is the fact that the agents, if kept separate till required, at the time of using produce a fresher test solution. Every one is familiar with the unstableness of solutions for the most part used in sugar testing. In fact this has been the great bar to perfecting a system of sugar testing up to this time; even Fehling's test becomes unreliable if kept too long.

Picric acid papers possess the advantage that they may be carried in the case and used in testing for both sugar and albumin. Used with the soda carbonate paper, the picric upon boiling with urine assumes a dark color if sugar be present.

The indigo carmine test, brought forward by Dr. Oliver last May, is the most striking and beautiful in reaction of any test with which I am familiar, and it is likely to prove the most reliable, both as a qualitative and quantitative test for sugar, of all. This test depends upon the fact first observed by Méhu, of Paris, in 1880, that when the carmine of indigo is treated with soda carbonate and a solution of glucose, or saccharine urine, the blue color is converted gradually into violet, then into various tints of red, and finally into yellow. Dr. Oliver has pointed out that the "indigo carmine and carbonate of soda in water, undergoes a gradual change, which renders the test unfit for use in that form. The test papers, however, not only meet these disadvantages, but amplify the powers of the test. The constituents being dry, remain unchanged; and when dissolved out of the papers, they furnish a freshly prepared solution at each observation." The method of using this test is as follows: Place in a test tube containing thirty minims of water, a paper charged with the carmine of indigo, and one of the soda carbonate. Gently heat till the indigo is discharged into the water, then add one or two drops (no more) of the suspected urine, and boil. If sugar be present, the blue quickly changes to a violet tint, this in turn, deepens and passes into purple, which next shades into reddish purple, followed by various tints of red; these quickly merge into orange-red, and orange, and at last the solution becomes of a straw color, which remains, however much boiled, and the paper now assumes the same yellow color.

The compass of this wonderful color reaction, embraces all the prismatic colors save green, and their kaleidoscopic play before the eye of the observer in testing, is one of most exquisite beauty.

It will be noted on standing the tube aside, that these colors slowly return, in the inverse order to which they appeared, especially in the upper portion of the tube, which is due to re-oxidation, through atmospheric contact. This may be hastened, as well as diffused through the whole tube, by continuous agitation. The tint reached in any particular case, depends upon the quantity of glucose present in the urine. It may stop at any of the colors reached, but be made to proceed to yellow by adding more glucose to the solution, and upon this fact depends the workings of the quantitative tests used in paper form.

WORKING TABLE.

POSITION IN POINT OF
SENSITIVENESS.

I.
Shows 1 part in 20,000.

Potassio mercuric
iodide.
Citric acid.

II.
A little less.

Sodium tungstate.
Citric acid.

III.
A little less.

Picric acid.
Citric acid.

IV.
Shows 1 part in 10,000.

Potassium ferrocyanide.
Citric acid.

ALBUMIN.

Of filtered urine take 30 minims, and drop in a citric acid paper, allowing a few moments for it to dissolve. If it clouds, it shows either:

URIC ACID, *dispelled by heat.*

MUCIN, *not dispelled by heat.*

OLEO-RESIN, *dispelled by heat, but reappearing on cooling.*

Filter if necessary, and to the clear fluid add one of the albumin reagents, which will cloud the liquid according to the amount present, rather increased by the application of heat.

A precipitate produced by the mercuric reagent or by picric acid, which redissolves on heating, indicates the presence of a vegetable alkaloid—generally quinine or cinchonidine.

I.
One drop of urine containing 1 per cent. of sugar will give a distinct reaction.

Indigo.
Sodium carbonate.

II.
Detects 1 part of sugar in 1,000 of fluid.

Picric acid.
Sodium carbonate.

SUGAR.

Into 30 minims of water in a test tube, place an indigo and a soda paper and one or two drops only of the suspected urine. At first you will have a pale blue solution, which on boiling, if sugar be present, will change to amethyst, and then to a pale yellow. On exposure to the air by shaking the tube, it goes back to blue. The indigo solution must be of only a pale blue color.

Proceed as above, using two or three soda papers instead of one (or, better still, a pinch, about three grains, of sodium carbonate). On boiling, sugar gives a dark-red or even brown color. Normal urine reacts slightly with this test.

URINARY TEST TABLETS.

SUPERIOR SUBSTITUTES FOR URINARY TEST PAPERS.

We now furnish (in tubes of 25 tablets of any one test) any of the following Urinary Tests in Tablet form:

CITRIC ACID.
INDIGO-AND-SODIUM-CARBONATE.
PICRIC ACID.
POTASSIUM FERROCYANIDE.

POTASSIO MERCURIC IODIDE.
SODIUM CARBONATE.
SODIUM TUNGSTATE.
BISMUTH SUBNITRATE (Böttcher's Sugar Test).

The Urinary Tests in paper form, especially those depending upon the production of a precipitate, have had one serious objection urged against them. If one of these Reagent Papers be boiled with a quantity of water, the agitation incident to ebullition is sufficient to disintegrate the paper, and the floating fibres are liable to be mistaken for a precipitate. In the substitution, for these papers, of tablets consisting of a soluble inert base combined with the reagent, we have not only obviated this defect, but have in several instances been enabled to simplify the tests, and, at the same time, enhance the permanency thereof.

The same general directions apply to the use of the *tablets* as for the papers.

The indigo test *paper* must be used in connection with one of the sodium carbonate *papers*, while the indigo *tablet* contains the sodium carbonate in combination with the indigo-carmin, thereby making it complete in itself. The quantity of sodium carbonate in the tablet is usually sufficient, but when very hard water is used to form the solution, or the urine is excessively acid, the addition of one of the sodium carbonate *tablets* becomes necessary.

The characteristic reaction which indicates the presence of glucose in the urine may be described as follows: Having dissolved one of the indigo and sodium carbonate tablets in about one drachm of water, the solution is heated to nearly the boiling point, and a drop of the urine is then added to the solution.

If sugar is present, a beautiful violet tint develops, which deepens and passes into purple; this gradually turns into reddish purple, which gives place to various tints of red, and these as quickly merge into orange red and orange, and finally the solution becomes of a straw color, which remains without further change. While observing the various changes of color which the liquid undergoes, any agitation of the solution should be carefully avoided. If the color of the solution remains a bright blue after the application of heat and the addition of one drop of urine, another drop of urine should be added, and the effect carefully noted. If after the addition of two or three drops of urine no change takes place, it is safe to conclude that sugar is not present in an abnormal quantity. If sugar is present, and the final yellow color has been developed, the various tints may be again produced in an inverse order by shaking the liquid. This remarkable effect is not due to cooling, but to the oxidizing influence of the air, and shows the importance of keeping the liquid as quiet as possible, after the addition of each drop of urine.

The ferrocyanide of potassium tablet furnishes a very reliable test for albumen. It differs, however, from the potassio-mercuric iodide test in that no precipitate is formed with peptone. In applying this test boiling must be avoided, otherwise decomposition of the ferrocyanide takes place.

BÖTTCHER'S SUGAR TEST IN TABLET FORM.

When boiled in the presence of an alkali, *bismuth subnitrate* is reduced to a suboxide, by grape sugar, the reduction being indicated by a change in the color of the bismuth, varying from a light yellow to black, according to the quantity of sugar present. So far as is known sulphur is the only substance likely to lead to a misrepresentation of results, and its presence in albuminous, bloody, or purulent urine is readily detected by the employment of lead oxide.

To 30 minims of the suspected urine, add a tablet of bismuth subnitrate, three tablets of sodium carbonate (a larger number if the urine be unusually acid) and boil for at least one minute. If now either sugar or sulphur be present, the bismuth will undergo a change of color, and in order to determine to which of these substances the reaction is attributable, in another portion of the urine substitute a tablet of litharge of bismuth, and proceed as before.

If the coloration in the previous test was occasioned by sulphur, the litharge will at once be reduced to a black sulphide, but if by sugar no change will result.

Physician's Vest Pocket Laboratory.

A COMPLETE SET OF APPARATUS AND REAGENTS FOR THE CLINICAL EXAMINATION OF THE URINE AT THE BEDSIDE.



REAGENT CASE WITH TEST TABLETS.

The reagent case with test papers contains the same apparatus as the case with tablets, together with the following test papers:

1. Litmus paper (neutral shade).
2. Reagents for Sugar.
 - (a) Picric Acid.
 - (b) Indigo Carmine.
3. Tests for Albumen.
 - (a) Potassio mercuric iodide.
 - (b) Sodium tungstate.
 - (c) Picric Acid.
 - (d) Potassium ferrocyanide.
4. Reagents used with the above.
 - (a) With the sugar tests, sodium carbonate.
 - (b) With the albumen tests, citric acid.

The above tests are as reliable as any others heretofore employed, while the information they afford is more precise and more varied in character. The graduated apparatus facilitates quantitative as well qualitative tests. Prices furnished upon application.

The pocket cases illustrated in the accompanying cuts enable the physician to carry with him the means for making all the qualitative and quantitative tests ordinarily required in examinations of the urine for clinical purposes. The reagent case with test tablets contains

the tests indicated in the opposite cut, and in addition the following apparatus:

- (1.) Two test tubes, one graduated.
- (2.) A graduated minim pipette.

(3.) A set of six specific gravity beads, which may be carried in the pocket without danger of breakage.

Also a little manual, giving complete instructions for the use of the tests; Vogel's scale of colors, and a card with the test lines employed by Dr. Oliver in quantitative estimations of albumen.



REAGENT CASE WITH TEST PAPERS.

PARKE, DAVIS & COMPANY,

Manufacturing Chemists,

DETROIT, MICHIGAN.

NEW YORK CITY: } 60 Maiden Lane,
 } 21 Liberty Street,
 } 218 Pearl Street.